LYSIS AND AGGLUTINATION OF RED CORPUSCLES OF WARM-BLOODED ANIMALS BY THE NORMAL SERUM OF COLD-BLOODED ANIMALS

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INTRODUCTION

In 1902 H. Noguchi[1] made extensive observations on the agglutination and solution of red corpuscles by the serum of different cold-blooded animals. He states that the serum of painted turtles have no lytic activity for the blood corpuscles of flat fish, congo eel, common sea eel, speckled turtle, and snapping turtle; slight activity for the blood corpuscles of bull-frog, and tautog; complete lytic activity for the blood of dog-fish; while agglutinins for many kinds of blood corpuscles are present in large quantities.

Since then, however, there have been relatively few reports on this problem (E. Friedberger and A. Seeling (Rana esculenta var ridibunda),[2] K. Landsteiner and Hans Rock (frog serum),[3] Liefmann,[4] Fränkel,[4a] and especially K. Mazzetti[4b]). Mazzetti tested serum of different species of reptiles and amphibians against the red corpuscles of calf. He divided the animals tested into 3 classes: (1) animals whose serum have no hemolytic activity—lizard and turtle; (2) animals whose serum have slight hemolytic power—frog and green lizard, and (3) animals whose serum exercise strong hemolysis—salamander, toad and snakes in general.

Testing the lytic action with red corpuscles of a few other warm-blooded animals, besides the cold-blooded animals mentioned, he found that the activity of lytic serum is more or less general against the red corpuscles of warm-blooded animals. He states that 0.1 c c of fresh normal serum of turtle, the specific name of which he does not give, has no lytic action on 1 c c of 5% suspension of red corpuscles of turtle, lizard, frog, toad, snake, guinea-pig, rabbit and calf.

[1] Bull. Univ. of Penn., 1902, 14, p. 438.
[2] Centralbl. f. Bakt., O., I, 1908, 46, p. 421.
[3] Ztschr. f. Immunitätsforsch., 1912, 14, p. 14.
[4] Berl. klin. Wchnschr., 37, 1900, p. 1682.
[4a] Ztschr. f. Immunitätsforsch., 1911, 10, p. 415.
[4b] Ibid., 1913, 18, p. 132.

In the course of my study[5] of the action of different serums on paramecia I observed the occurrence of lysins in the serum of the painted turtle (Chrysemys picta) against the blood of rabbit and few other warm-blooded animals. This fact seems to be in contradiction to the statement of Mazzetti.

In this paper the hemolysins and hemagglutinins of the turtle serum will be discussed.

TABLE 1

TESTS TO DETERMINE LYSIS AND AGGLUTINATION OF TURTLE SERUM

	1	2	3	4	5	6	7
Dilution of the turtle serum with 0.85 % NaCl solution to 1 : 5	0.5	0.4	0.3	0.2	0.15	0.1	—
0.85 % NaCl solution	—	0.1	0.2	0.3	0.35	0.4	0.5
5 % suspension of red corpuscles of different kinds of animals in 0.85 % NaCl solution	0.5	0.5	0.5	0.5	0.5	0.5	0.5
After 3 hours at room temperature:							
Human—							
Lysis	+++	+++	+++	+	±	—	—
Agglutination	+++	+++	+++	+++	—	—	—
Hog—							
Lysis	+++	+++	+++	+++	+++	+	—
Agglutination	+++	+++	+++	+++	+++	+++	—
Calf—							
Lysis	+++	++	+	—	—	—	—
Agglutination	+++	+++	+++	+++	—	—	—
Guinea-pig—							
Lysis	—	—	—	—	—	—	—
Agglutination	+++	+++	+++	+++	—	—	—
Rabbit—							
Lysis	+++	+++	+++	+++	+++	+++	—
Agglutination	+++	+++	+++	+++	+++	+++	—
Pigeon—							
Lysis	—	—	—	—	—	—	—
Agglutination	+++	+++	++	—	—	—	—
Sheep—							
Lysis	++	+	—	—	—	—	—
Agglutination	+++	++	—	—	—	—	—

NORMAL HEMOLYSIS AND HEMAGGLUTININS IN THE SERUM OF THE PAINTED TURTLE (CHRYSEMYS PICTA) AND OTHER SPECIES

All turtles were obtained in the beginning of March and kept in a box with some tap water at room temperature of the animal house of this laboratory. They were therefore in the condition of their hibernation when they were bled.

The turtle serum is said to have no, or at least not an appreciable amount of, hemolysins against different kinds of red corpuscles of cold-blooded animals (Noguchi, Mazzetti), as well as against the corpuscles of guinea-pig, rabbit and calf (Mazzetti).

Testing the normal serum of Chrysemys picta, I found that this serum contains an appreciable amount of hemolysins for the red corpuscles of different warm-blooded animals, besides normal agglutinins against different kinds of

[5] Takenouchi: Jour. Infect. Dis., 1918, 23, p. 396.

red corpuscles of cold-blooded animals. I tested the red corpuscles of the following animals to determine their reaction to agglutination and dissolution by normal turtle serum.

Human: positive lysis and agglutination.
Calf: positive lysis and agglutination.
Sheep: slightly positive lysis and agglutination.
Hog: strong positive lysis and agglutination.
Rabbit: strong positive lysis and agglutination.
Guinea-pig: negative lysis and positive agglutination.
Pigeon; no lysis and positive agglutination.

As is usual when biologic laking agents act on the red corpuscles of different kinds of animals, the hemolytic agent of the normal turtle serum, even in quantities more than sufficient to liberate all the hemoglobin, does not destroy the structure of the erythrocytes. Agglutination takes place, in almost all cases, quicker than the lysis; hemoglobin is generally liberated from the red corpuscles which are already agglutinated in more or less marked degree.

TABLE 2

SUMMARY OF RESULTS OF TESTS WITH DIFFERENT SPECIES OF TURTLES

	1 Chrysemys picta	2 Chrysemys belli	3 Chrysemys elegans	4 Melaco-clemmys lesueurii	5 Melaco-clemmys (geograph-ica?)
Human—					
Lysis	0.2				
Agglutination	0.2				
Hog—					
Lysis	0.1	0.2	0.15	0.2	0.3
Agglutination	0.1	0.15	0.15	0.1	0.4
Calf—					
Lysis	0.4	—	0.5	—	—
Agglutination	0.2	0.3	0.15	0.4	—
Guinea-pig—					
Lysis	—	0.3	0.15	0.4	0.4
Agglutination	0.2	0.2	0.15	0.15	0.4
Rabbit—					
Lysis	0.1	0.1	0.1	0.15	0.2
Agglutination	0.1	0.1	0.1	0.1	0.3
Pigeon—					
Lysis	—	—	—	—	—
Agglutination	0.3	0.3	0.1	0.3	—

After the complete lysis has taken place, the ghosts are also agglutinated into many flakes or into a compact mass or have the appearance of a membrane floating in the hemoglobin solution. Using the terminology of Stewart,[6] the process of hemolysis by the turtle serum is really hemochromolysis and not hemocytolysis.

Several other different species of turtles, Chrysemys belli, Chrysemys elegans, Melacoclemmys lesueurii, and Melacoclemmys (geographica?), were tested with the same method on hemolysins and hemagglutinins, and the results summarized in Table 2. The titer limits of hemolysis and agglutination with different kinds of blood corpuscles are given in amount of serum dilution of 1:5.

The serum of different species of normal turtle contains different amounts of hemolysins and hemagglutinins for corpuscles of different kinds of animals.

[6] Jour. Pharmacol. and Exper. Therap., 1909, I, p. 89.

INFLUENCE OF LOW TEMPERATURE ON THE HEMOLYTIC AND HEMAGGLUTINATING PROCESS BY THE NORMAL TURTLE SERUM

It is well known that temperature plays an important part in the development of hemolysis. At a temperature approaching freezing it is usually absent in serum of warm-blooded animals. Agglutination, on the other hand, is not prevented by low temperature (Flexner[7]). Friedberger and Seeling[8] observed that the serum of the frog, unlike the serum of warm-blooded animals, causes complete hemolysis at 0 C., tho at a much slower rate.

The relation between the temperature and the readiness with which the lysis and agglutination of red corpuscles of rabbit blood succumb to the effects of turtle serum, will be seen in the following experiment.

TABLE 3

EFFECT OF TEMPERATURE ON LYSIS AND AGGLUTINATION

	1	2	3	4	5	6	7
Serum of Chrysemys picta diluted with 0.85 % NaCl to 1 : 10	0.5	0.4	0.3	0.2	0.15	0.1	—
0.85 % NaCl solution	—	0.1	0.2	0.3	0.35	0.4	0.5
5 % suspension of rabbit corpuscles	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Lysis at 23 - 24 C.:							
15 minutes	+++	+++	+++	—	—	—	—
20 minutes	+++	+++	+++	++	+	—	—
60 minutes	+++	+++	+++	+++	+++	±	—
Lysis at 7 C.:							
2 hours	+++	—	—	—	—	—	—
3 hours	+++	+++	—	—	—	—	—
Lysis at 1 - 5 C.:							
2 hours	+*	—*	—*	—*	—*	—	—
3 hours	+++	++	—	—	—	—	—

* The agglutination of the corpuscles takes place in 1 hour in the first 5 tubes of the last series (at a temperature of 1 - 1.5 C.) in more or less marked degree.

Testing with 1 : 5 dilution of the serum of Chrysemys picta, we get the following result:

TABLE 4

EFFECT OF TEMPERATURE WHEN A DIFFERENT DILUTION IS USED

	1	2	3	4	5
Turtle Serum 1 : 5	0.5	0.4	0.3	0.2	—
0.85 % NaCl	—	0.1	0.2	0.3	0.5
5 % suspension of rabbit corpuscles	0.5	0.5	0.5	0.5	0.5
Temperature: Time:					
At 15-17 C. 3 minutes, lysis	+++	+++	+++	++	—
8-8.5 C. 15 minutes, lysis	±	—	—	—	—
20 minutes, lysis	+++	-	—	—	—
0-1.0 C. 27 minutes, lysis	+	—	—	—	—
40 minutes, lysis	+++	++	+	—	—

[7] Bull. Univ. of Penn., 1902, 15, p. 324.

[8] Centralbl. f. Bakteriol., O., 1, 1908, 46, p. 421.

ACTION OF TURTLE SERUM ON HARDENED CORPUSCLES

It is stated[9] that red corpuscles which have been hardened in Hayem's fluid are still lysable by lytic serums. Noguchi[10] tested the action of agaricin, saponin, and tetanolysis on the red corpuscles of man, guinea-pig, and rabbit, hardened in Hayem's solution, formalin, and alcohol-ether, but did not observe any solution of the red cells or liberation of hemoglobin. On the other hand, corpuscles hardened in Hayem's solution and formalin are still capable of agglutination with ricin and the venom of cobra, moccasin, copperhead and rattlesnakes as well as with normal horse serum. Von Dungern and Coca[11] announced that osmic-hardened corpuscles can be laked by foreign serum. According to Stewart, sublimate fixed corpuscles at a certain stage of fixation

TABLE 5

TESTS TO DETERMINE THE ACTION OF TURTLE SERUM ON HARDENED CORPUSCLES

		1	2	3	4	5	'6	7
Active normal serum of Chrysemys picta, diluted to 1:5 with 0.85 % NaCl solution		0.5	0.4	0.3	0.2	0.15	0.1	—
0.85 % NaCl solution		—	0.1	0.2	0.3	0.35	0.4	0.5
5 % suspension of hardened rabbit corpuscles in 0.85 % NaCl solution		0.5	0.5	0.5	0.5	0.5	'0.5	0.5
Time of Fixation in Hayem's Solution:	Time of Observation:							
One hour	15 mins.							
	Lysis	—	—	—	—	—	—	—
	Agglutination	++	++	+	+	—	—	—
	40 mins.							
	Lysis	—	—	—	—	—	—	—
	Agglutination	++	++	++	+	+	—	—
	1½ hr.							
	Lysis	+	+	±	—	—	—	—
	Agglutination	++	++	++	+	+	—	—
	20 hr.							
	Lysis	+	+	+	—	—	—	—
	Agglutination	++	++	++	++	+	+	–
Two hours	15 mins.							
	Lysis	—	—	—	—	—	—	—
	Agglutination	++	++	+	+	—	—	—
	1 hr.							
	Lysis	—	—	—	—	—	—	—
	Agglutination	++	++	++	+	+	—	—
	20 hr.							
	Lysis	—	—	—	—	—	—	—
	Agglutination	++	++	++	+	+	+	—
Three hours*								

* No hemolysis took place in any tube; the degree of agglutination is just the same as above (2 hours' fixation).

do not lake in distilled water at room temperature, but lake in ammoniacal water, and are also laked in distilled water on heating. In blood partially fixed by formaldehyd and laked by saponin the ghosts are not broken up by water, as in ordinary blood laked by saponin.[8]

It was found that the hardened corpuscles of rabbit blood cannot be dissolved by the normal turtle serum (Chrysemys picta), but are agglutinated just as readily as the original blood corpuscles.

[9] Matthes: München. med. Wchnschr., 1902, 49, p. 8.

[10] Bull. Univ. of Penn., 1902, 15, p. 327.

[11] Berl. klin. Wchnschr., 1907, 44, p. 1471.

Blood, taken from a normal rabbit directly into the Hayem's solution, was placed at room temperature for different lengths of time until the corpuscles were centrifugalized repeatedly and washed thoroughly with physiologic salt solution.

The solution of corpuscles, or rather the liberation of hemoglobin, which took place in the first series was due to insufficient fixation.

CONCENTRATION OF ELECTROLYTES AND AGGLUTINATION OF HARDENED CORPUSCLES

In the action of the agglutinin on hardened corpuscles of rabbit blood I noticed that a certain salt concentration is invariably necessary for the agglutination of hardened corpuscles—a fact which agrees with that of the agglutination of different bacteria by specific or normal agglutinins.

TABLE 6

SALT CONCENTRATION NECESSARY IN THE ACTION OF AGGLUTININ ON HARDENED CORPUSCLES

		1	2	3	4	5	6
Turtle serum, diluted with 0.85 % NaCl solution to 1 : 5		0.3	0.3	0.3	0.3	0.3	0.3
Distilled water		—	0.1	0.2	0.3	0.4	0.5
0.85 % NaCl solution		0.5	0.4	0.3	0.2	0.1	—
10 % suspension of hardened rabbit corpuscles in water		0.2	0.2	0.2	0.2	0.2	0.2
Time of Observation:							
15-18 minutes	Agglutination	+++	+++	++	+	+	—
30 minutes	Agglutination	+++	+++	+++	++	+	—
20 hours	Agglutination	+++	+++	+++	+++	++	—

THE INACTIVATION OF NORMAL SERUM OF CHRYSEMYS PICTA

The heat lability of normal hemolytic complement in the serum of Chrysemys picta was determined. The serum was heated for 30 minutes to a temperature varying from 45-60 C., after which the lysis was again tested. Heating at 50 C. for 30 minutes is sufficient for complete inactivation of complement in the serum of Chrysemys picta, while heating at 45 C. for 30 minutes left the complement almost intact.

THE INACTIVATION OF TURTLE SERUM BY DIGESTION WITH DIMINISHED SALT CONCENTRATION (SACHS AND TERUUCHI)[12]

The digestion method was used with active fresh as well as somewhat old serums of the turtle, and the results were similar to those observed by Sachs and Teruuchi in their study with the serums of warm-blooded animals, namely, (1) the normal active turtle serum, which has been kept over 48 hours in an ice-box, can be inactivated when it is digested with water in a dilution of 1 : 9, but not of 1 : 5 (the dilution of the serums with water to 1 : 9-10 is said to be most favorable for inactivation); (2) fresh turtle serum cannot be inactivated by this method at all.

[12] Berl. klin. Wchnschr., 1907, 16, p. 467; 17, p. 520; 19, p. 602; and Tsuda, Ibid., 1908, 8, p. 399.

REACTIVATION OF INACTIVE TURTLE SERUM

Reactivation of inactivated normal turtle serum was attempted many times with normal complement of guinea-pig and of frog, but no reactivation was observed in either case.

HEAT LABILITY OF HEMAGGLUTININS IN THE SERUM OF CHRYSEMYS PICTA

Noguchi[18] showed that the agglutinins in the serum of Limulus polyphemus have varying degrees of heat lability, altho temperatures of 40 C., when continued for 30 minutes, diminish the activity of all the agglutinins. Temperatures approaching 65 C. seem to destroy wholly the agglutinating power of the serum for erythrocytes.

The results of my experiments with the inactive serum of Chrysemys picta and different kinds of red corpuscles of warm-blooded animals are as follows:

(*a*) The agglutinins for hog, rabbit and pigeon corpuscles do not show any marked diminution of activity on heating at 50 C. for 30 minutes or at 53-54 C. for 15 minutes. Agglutinin for rabbit corpuscles in the normal turtle serum is not entirely destroyed by heating the serum even at 60 C. for 30 minutes, when the serum becomes almost coagulated.

(*b*) The agglutinins for calf and sheep corpuscles seem to be entirely destroyed by heating at 50 C. for 30 minutes. The agglutinin for guinea-pig corpuscles in the turtle serum is destroyed to a great extent by heating the serum at 53 C. for 15 minutes.

The highest temperature at which the agglutinins in the normal turtle serum could be completely destroyed was not determined. In this experiment the agglutinins in the serum of Chrysemys picta are multiple in their nature; in other words, the agglutinins for some kinds of red corpuscles of warm-blooded animals are different from those for other corpuscles. The question regarding the multiplicity of the serum agglutinins of cold-blooded animals is answered by Noguchi in the affirmative. According to the result of my experiment on absorption the conclusion that the serum of Chrysemys picta contains several kinds of agglutinins seems most likely to be correct.

ABSORPTION TESTS

Fresh normal active serum of Chrysemys picta was diluted to 1:5 with 0.85% salt solution; to each 10 c c of this dilution 0.7 c c of red corpuscles of different kinds of animals is added. Each tube is placed in ice water at a temperature between 0 C.-1.5 C. for different lengths of time (from 10-17 minutes) and then centrifugalized. The supernatant fluid of each serum dilution and blood corpuscles is tested with a different kind of corpuscle for agglutination and lysis.

To avoid hemolysis during the absorption, the time of absorption must be controlled carefully, and the temperature must be kept low enough to make the rate of absorption of complement as slow as possible. The method of inactivating the serum by heating should not be employed here because normal hemagglutinins for some kinds of corpuscles are very heat labile.

[18] Centralbl. f. Bakteriol., 1903, O., I, 34, p. 286.

(*a*) Absorption of turtle serum (Chrysemys picta) by the red corpuscles of human blood.

Turtle serum (1:5) absorbed by the red corpuscles of human blood for 7 minutes at 0-1.5 C.; 3 minutes for centrifugalization.

TABLE 7*

SHOWING THE ABSORPTION OF TURTLE SERUM BY RED CORPUSCLES OF HUMAN BLOOD

Supernatant	0.5	0.4	0.3	0.2	0.1	—
0.85 % NaCl	—	0.1	0.2	0.3	0.4	0.5
5 % suspension of corpuscles	0.5	0.5	0.5	0.5	0.5	0.5
a. Human—						
Lysis	±	—	—	—	—	—
Agglutination	+	—	—	—	—	—
b. Calf—						
Lysis	—	—	—	—	—	—
Agglutination	+++	+++	+++	—	—	—
c. Hog—						
Lysis	+++	+++	+++	+++	+	—
Agglutination	+++	+++	+++	+++	—	—
d. Rabbit—						
Lysis	+++	+++	+++	+++	+++	—
Agglutination	+++	+++	+++	+++	+++	—
e. Hardened Rabbit—						
Lysis	—	—	—	—	—	—
Agglutination	+++	+++	+++	+++	+++	—

* Many readings were made at different intervals of time, but only the one at the 6th hour will be given in Tables 7-11.

(*b*) Absorption by the red corpuscles of rabbit blood; 7 minutes for absorption and 3 minutes for centrifugalization.

TABLE 8

ABSORPTION TESTS USING RABBIT BLOOD

Supernatant	0.5	0.4	0.3	0.2	0.1	—
0.85 % NaCl	—	0.1	0.2	0.3	0.4	0.5
5 % suspension of corpuscles	0.5	0.5	0.5	0.5	0.5	0.5
a. Human—						
Lysis	++	—	—	—	—	—
Agglutination	+	—	—	—	—	—
b. Calf—						
Lysis	—	—	—	—	—	—
Agglutination	+	±	—	—	—	—
c. Hog—						
Lysis	++	+	—	—	—	—
Agglutination	++	—	—	—	—	—
d. Rabbit—						
Lysis	+++	+++	+++	+	—	—
Agglutination	+++	+++	+++	+++	+	—
e. Hardened Rabbit—						
Lysis	—	—	—	—	—	—
Agglutination	+++	+++	+++	+++	—	—

From the results obtained in the absorption experiments described, the following statements seem warranted, though the time for the absorption was not sufficient in some cases because it was necessary to avoid any hemolysis which might occur during the absorption.

The result is somewhat different from that which was obtained by

(*c*) Absorption by the red corpuscles of hog blood; 7 minutes for absorption, 3 minutes for centrifugalization.

TABLE 9

ABSORPTION TESTS USING HOG BLOOD

Supernatant	0.5	0.4	0.3	0.2	0.1	—
0.85 % NaCl	—	0.1	0.2	0.3	0.4	0.5
5 % suspension of corpuscles	0.5	0.5	0.5	0.5	0.5	0.5
a. Human—						
Lysis	++	—	—	—	—	—
Agglutination	++	+	—	—	—	—
b. Calf—						
Lysis	—	—	—	—	—	—
Agglutination	+	+	—	—	—	—
c. Hog—						
Lysis	—	—	—	—	—	—
Agglutination	+++	++	+	—	—	—
d. Rabbit—						
Lysis	+++	+++	+++	+++	++	—
Agglutination	+++	+++	+++	+++	+	—
e. Hardened Rabbit—						
Lysis	—	—	—	—	—	—
Agglutination	+++	+++	+++	+++	+++	—

(*d*) Absorption by the red corpuscles of calf blood for 17 minutes and centrifugalization for 3 minutes.

TABLE 10

ABSORPTION TESTS USING CALF BLOOD

Supernatant	0.5	0.4	0.3	0.2	0.1	—
0.85 % NaCl	—	0.1	0.2	0.3	0.4	0.5
5 % suspension of corpuscles	0.5	0.5	0.5	0.5	0.5	0.5
a. Human—						
Lysis	+	—	—	—	—	—
Agglutination	+	—	—	—	—	—
b. Calf—						
Lysis	—	—	—	—	—	—
Agglutination	+	—	—	—	—	—
c. Hog—						
Lysis	+++	++	+	+	—	—
Agglutination	++	++	+	+	—	—
d. Rabbit—						
Lysis	+++	+++	+++	+++	—	—
Agglutination	+++	+++	+++	+++	+++	—
e. Hardened Rabbit—						
Lysis	—	—	—	—	—	—
Agglutination	+++	+++	+++	+++	—	—

Noguchi in his study of hemagglutinins in the serum of Limulus polyphemus against the different kinds of corpuscles of cold-blooded animals, where the absorption of the agglutinin for one species left those for others in practically undiminished quantities.

The lysins and agglutinins for hog corpuscles can be absorbed by the red corpuscles, not only of hog blood, but also of rabbit and of calf blood, tho not by human blood corpuscles.

(*e*) Absorption by hardened rabbit corpuscles for 20 minutes; 3 minutes for centrifugalization.

TABLE 11

SHOWING THE ABSORPTION BY HARDENED RABBIT CORPUSCLES

Supernatant	0.5	0.4	0.3	0.2	0.1	—
0.85 % NaCl	—	0.1	0.2	0.3	0.4	0.5
5 % suspension of corpuscles	0.5	0.5	0.5	0.5	0.5	0.5
a. Human—						
Lysis	+++	++	++	+	—	—
Agglutination	+++	+	+	—	—	—
b. Calf—						
Lysis	+++	+++	++	—	—	—
Agglutination	+++	+	±	—	—	—
c. Hog—						
Lysis	+++	+++	+++	++	+	—
Agglutination	+++	+++	++	—	—	—
d. Rabbit—						
Lysis	+++	+++	+++	+++	+++	—
Agglutination	+++	+++	+++	+++	++	—
e. Hardened Rabbit—						
Lysis	—	—	—	—	—	—
Agglutination	+++	++	+	—	—	—

The lysins and agglutinins for rabbit corpuscles and the agglutinins for hardened corpuscles of rabbit blood are absorbed most strongly by the corpuscles of the same animals and cannot be absorbed very much by other kinds of corpuscles.

The lysins and agglutinins for human corpuscles are almost equally absorbed by human, rabbit, calf and hog corpuscles.

The lysins for calf blood corpuscles can be absorbed by different kinds of corpuscles.

The hardened corpuscles of rabbit blood cannot absorb very much any kind of lysins in the turtle serum.

RÉSUMÉ

The normal serum of different species of turtle contain different amounts of lysins and agglutinins for different kinds of corpuscles of warm-blooded animals.

The normal serum of turtle (Chrysemys picta) contains several lysins and agglutinins for erythrocytes of warm-blooded animals.

The normal lysins of the turtle serum lake the red corpuscles of warm-blooded animals even at freezing temperature.

The red corpuscles of rabbit blood hardened by Hayem's solution cannot be laked by the turtle serum, unless the fixation is incomplete.

For the agglutination of hardened corpuscles a certain concentration of electrolyte is necessary.

Reactivation of inactive turtle serum by normal complement of guinea-pig or frog was tested with negative result.

The agglutinins of turtle serum for erythrocytes of warm-blooded animals have varying degrees of heat lability.

Red corpuscles of some species of the tested animals can absorb agglutinins and lysins for erythrocytes of other animals. The specificity of normal agglutinins and lysins in turtle serum (Chrysemys picta) for erythrocytes of warm-blooded animals seems not to be so strict as claimed by Noguchi in the serum of Limulus polyphemus and Musterus canis for erythrocytes of different cold-blooded animals.

However, the serum of turtle (Chrysemys picta) seems to contain not only a multiplicity of agglutinins, but also a multiplicity of lysins for red corpuscles of warm-blooded animals.